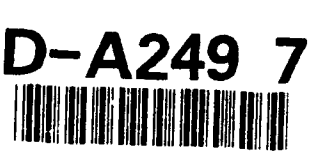

FINAL TECHNICAL REPORT FOR OFFICE OF NAVAL RESEARCH GRANT
N00014-90-J-1663 FROM 1 DECEMBER 1989 TO 31 MAY 1991

PRINCIPAL INVESTIGATOR: RALPH MITCHELL
DIVISION OF APPLIED SCIENCES, HARVARD UNIVERSITY
CAMBRIDGE, MASSACHUSETTS 02138

SUMMARY OF WORK ACCOMPLISHED

This grant supported basic research in a project involving the study of interactions between marine bacteria and various substrata during the process of bacterial adhesion and was a continuation of a previous contract. During this period, one manuscript was published, one manuscript accepted for publication, one manuscript submitted for publication and under review, with two further manuscripts in preparation. The titles of these publications/manuscripts and their disposition are listed at the end of this report.

The results obtained during the course of this project on substratum-microorganism interactions and the adhesion of marine bacteria are summarized as follows:

(1) A series of field experiments indicated that there are substratum influences on the attachment of bacteria in Antarctic marine waters. Detachment of bacteria from the substrata also appeared to occur. (see Maki, Little, Wagner & Mitchell 1990)

(2) Some bacteria, when in suspension, were demonstrated to have different cell surface hydrophobicities using the adhesion to hexadecane technique. However, by measuring contact angles of both air bubbles and hexadecane droplets of resulting films of these same bacteria, the film wettability was determined to be the same. (see Maki, Rittschof & Mitchell 1992)

(3) Our observations of a copiotrophic marine bacterium under conditions of nutrient deprivation showed that it underwent fragmentation (i.e., cell division without growth) and formed dwarf cells with an increase in cell surface hydrophobicity. The bacterium at the initiation of nutrient deprivation conditions more easily attached to a wettable substratum (i.e., glass) than to a non-wettable substratum (i.e., polyvinylchloride, PVC). After 24 hours of nutrient deprivation, adhesion to the two substrata was not different.

(4) Protein synthesis continues in cells undergoing a starvation response. Chloramphenicol was used to block protein synthesis in starved cells (demonstrated by blocking the incorporation of ^{3}H-leucine into trichloroacetic acid insoluble material). The inhibition of protein synthesis on cells affected their adhesion to PVC but not glass at the initiation of nutrient deprivation. However, after 24 hours no effect on adhesion to either substrata was observed.

(5) Conjugation between enteric bacteria and marine bacteria resulted in the insertion of a transposon (mini-Mu) into the genome of the marine bacteria. Electron microscopy showed that the mutants possessed extracellular polymers but analyses using calcofluor white and lectins indicated that these polymers were different from the polymers of the wild-type. This change in polymers also affected the ability of these cells to firmly attach to various substrata.

PUBLICATIONS INVOLVING THIS GRANT 1989-1991

(1) Maki JS, Little BJ, Wagner P, Mitchell R. 1990. Biofilm formation on metal surfaces in Antarctic waters. Biofouling 2: 27-38.

(2) Maki JS, Rittschof D, Mitchell R. 1992. Inhibition of larval barnacle attachment to bacterial films: an investigation of physical properties. Microbial Ecology (in press)

(3) Maki JS, Samuelsson M-O, Rittschof D, Szewzyk U, Kjelleberg S, Mitchell R. Substratum/bacterial interactions and their effect on the attachment of *Balanus amphitrite* cypris larvae. Manuscript in Review.

(4) Maki JS, Yule AB, Rittschof R, Mitchell R. Effect of bacterial films on the temporary adhesion of cypris larvae, *Balanus amphitrite* Darwin. Mauscript in Preparation.

(5) Maki JS, Joyce EA, Mitchell R. The effect of inhibition of protein synthesis on the adhesion of a copiotrophic bacterium during nutrient deprivation. Manuscript in Preparation.

Statement A per telecon
Dr. Bernard Zahuranec ONR/Code 1123
Arlington, VA 22217-5000

NWW 5/7/92